

CLEAN VERSION OF AMENDED CLAIMS - OZ 50531

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4. An electron donor system as claimed in claim 1, wherein the mediator has a standard normal potential in the region of less than about -0.4 V.
 5. An electron donor system as claimed in claim 1, wherein the mediator is selected from cobalt(III) sepulchrate, methylviologen, neutral red, riboflavin, ruthenium triacetate, FMN and FAD.
 6. An electron donor system as claimed in claim 1, wherein the source of electrons is a metal with a lower standard normal potential than the mediator.
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8. An electron donor system as claimed in claim 1, selected from the systems:
- Zn/cobalt(III) sepulchrate and
 - Zn/neutral red.
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9. A method for the enzymatic transfer of oxygen to a hydrocarbon-containing hydrogen donor molecule, which comprises incubating the hydrogen donor molecule in a reaction medium comprising the oxygen-transferring enzyme and an electron donor system as claimed in claim 1 in the presence of oxygen under reaction conditions.
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11. A method for the enzymatic production of terminally or subterminally (position ω -1 to ω -4) hydroxylated fatty acids, which comprises
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- a) converting a hydroxylatable fatty acid or fatty acid derivative in the presence of an electron donor system as claimed in claim 1 using a cytochrome P450 mono oxygenase and oxygen; and
 - b) isolating the hydroxylated product(s).

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13. A method as claimed in claim 9, wherein the enzyme is a cytochrome P450 mono oxygenase selected from:

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- a) the wild-type enzyme which can be isolated from *Bacillus megaterium* (DSM 32T); or
 - b) a mutant, which can be obtained by amino acid substitution in at least one of positions 26, 47, 72, 74, 87, 188 and 354, of the wild-type enzyme (SEQ ID NO: 35).
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16. A method as claimed in claim 11, wherein the electron donor system is zinc/Co(III) sepulchrate.

17. A method as claimed in claim 11, wherein at least stage a) is carried out in the presence of chloride ions.

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18. A method as claimed in claim 11, wherein at least stage a) is carried out in the presence of a hydrogen peroxide-cleaving enzyme.

19. A bioreactor for use for producing ω -hydroxylated fatty acids, which comprises immobilized monooxygenase and an electron donor system as claimed in claim 1 in a liquid reaction medium.

20. A detection method for fatty acid monooxygenases, which comprises

- a) incubating an analyte suspected of having enzymic activity with an ω -hydroxylatable fatty acid or fatty acid derivative which has a terminal chromophore or fluorophore which can be eliminated, in the presence of an electron donor system as claimed in claim 1; and

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- b) determining the elimination of the chromophore or fluorophore
qualitatively or quantitatively.
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22. A test kit comprising an electron donor system as claimed in claim 1.

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4. An electron donor system as claimed in claim 1 [any of the preceding claims], wherein the mediator has a standard normal potential in the region of less than about -0.4 V.
5. An electron donor system as claimed in claim 1 [any of the preceding claims], wherein the mediator is selected from cobalt(III) sepulchrates, methylviologen, neutral red, riboflavin, ruthenium triacetate, FMN and FAD.
6. An electron donor system as claimed in claim 1 [any of the preceding claims], wherein the source of electrons is a metal with a lower standard normal potential than the mediator.
8. An electron donor system as claimed in claim 1 [any of the preceding claims], selected from the systems:
 - Zn/cobalt(III) sepulchrates and
 - Zn/neutral red.
9. A method for the enzymatic transfer of oxygen to a hydrocarbon-containing hydrogen donor molecule, which comprises incubating the hydrogen donor molecule in a reaction medium comprising the oxygen-transferring enzyme and an electron donor system as claimed in claim 1 [any of claims 1 to 8] in the presence of oxygen under reaction conditions.
11. A method for the enzymatic production of terminally or subterminally (position ω -1 to ω -4) hydroxylated fatty acids, which comprises
 - a) converting a hydroxylatable fatty acid or fatty acid derivative in the

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- presence of an electron donor system as claimed in claim 1 [any of claims 1 to 8] using a cytochrome P450 mono oxygenase and oxygen; and
- b) isolating the hydroxylated product(s).
13. A method as claimed in claim 9 [any of claims 9 to 12], wherein the enzyme is a cytochrome P450 mono oxygenase selected from:
- a) the wild-type enzyme which can be isolated from *Bacillus megaterium* (DSM 32T); or
- b) a mutant, which can be obtained by amino acid substitution in at least one of positions 26, 47, 72, 74, 87, 188 and 354, of the wild-type enzyme (SEQ ID NO: 35).
16. A method as claimed in claim 11 [any of claims 11 to 15], wherein the electron donor system is zinc/Co(III) sephulchrute.
17. A method as claimed in claim 11 [any of claims 11 to 16], wherein at least stage a) is carried out in the presence of chloride ions.
18. A method as claimed in claim 11 [any of claims 11 to 17], wherein at least stage a) is carried out in the presence of a hydrogen peroxide-cleaving enzyme.
19. A bioreactor for use for producing ω -hydroxylated fatty acids, which comprises immobilized monooxygenase and an electron donor system as claimed in claim 1 [any of claims 1 to 8] in a liquid reaction medium.
20. A detection method for fatty acid monooxygenases, which comprises

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- a) incubating an analyte suspected of having enzymic activity with an ω -hydroxylatable fatty acid or fatty acid derivative which has a terminal chromophore or fluorophore which can be eliminated, in the presence of an electron donor system as claimed in claim 1 [any of claims 1 to 8]; and
 - b) determining the elimination of the chromophore or fluorophore qualitatively or quantitatively.
22. A test kit comprising an electron donor system as claimed in claim 1 [any of claims 1 to 8].

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